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RESPONSE OF RICE TO FERTILIZATION IN THE SEEDBEDS 1/

A. A. BRIONES, J. G. DAVIDE, and N. L. GALVEZ

Seedbed fertilization of rice, Milfor 6 (2), did not affect the different growth characteristics like height, panicle, and tiller counts at maturity but significantly increased the grain yield. This increase was obtained by the addition of nitrogen or potassium in the seedbeds. There was no consistent response to phosphorus.

Grain yields were always higher in the "dapog"- than in the "punla"-raised seedlings over all seasons. The yield of straw was not affected by fertilizer addition.

As the growing need for the use of fertilizers and the high cost of fertilization are felt, the wise use of fertilizers should be explored to obtain maximum efficiency. In paddy rice culture, different methods of fertilizer application have been found to give higher yields than most unfertilized lots. In Japan, for instance, deep placement of ammonium sulfate has been found to increase the yields 2 1/2 times that of the control and 10 per cent greater than similar rates of the fertilizer applied on the surface of the paddies (De Geus, 1954). Split application (i.e., applying a part at planting and the rest before booting) of nitrogenous fertilizer to lowland rice usually increases the yield of rice, and is generally better than one application at any stage of growth. In India, where extensive fertilization studies in lowland rice are being conducted, results showed responses to application of nitrogen and phosphorus but rarely to potash fertilizers (De Geus, 1954).

Although seedbed fertilization cannot substitute for fertilization in the fields of low levels of soil fertility, this practice may augur sturdy and vigorous seedlings which can stand better the drastic environmental changes brought about during transplanting.

Farmers usually grow seedlings in the seedbeds before transplanting. These seedlings invariably exhibit chlorosis attributable to nitrogen deficiency as a result of thick sowing and/or low seedbed fertility. The deficiency is virtually compounded when the

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1/ Central Experiment Station Contribution No. 2824. Part of Rice and Corn Corn Research Project No. 6.

seedlings are transplanted to paddies of low fertility.

Velasco *et al.* (1953) obtained results indicating that rice plants need ammonium sulfate at the seedling, at mid-vegetative, and at the early reproductive stages. Espino (1928) estimated that only about 53 per cent of the early growth and development of young rice plants is supported by the food supplied by the seed.

Songcuya (1941), working on Elon-elon rice variety, found that the different amounts of 9-9-4 mixed fertilizer applied in the seedbed alone, in both the seedbed and the field, or in the field alone did not materially influence the flowering and maturity periods and the number of bearing culms per hill. In general, the plants fertilized in the seedbed gave lower yields than those fertilized in the field. In his study, however, no actual transplanting was done. The germinated seeds were merely sown in the puddled paddy ("palatak" system) and the first two weeks was considered as the hypothetical seedbed stage.

The pre-treatment of seeds also received considerable attention from several rice workers. De Geus (1954) cited the works in Ceylon and other countries which showed significant yield increases in pot experiments using seeds soaked in 5 and 20 per cent Na_2HPO_4 and K_2HPO_4 solutions. Unfortunately, these increases in yield were not reproduced under field conditions. In India, he reported that soaking seeds in 10 and 20 per cent solutions of K_3PO_4 resulted in significant yield increases of 21 and 30 per cent, respectively, over the control.

Homans (1952) observed that it was possible to increase yields by about 500 kg. per hectare through the fertilization of seedbeds and the crop in the field as compared to fertilization of the crop in the field alone. Van de Goor (1951) showed that quantities of phosphorous absorbed during the seed treatment were not sufficient to warrant omission of the usual soil treatment. Likewise, he noted that the application of fertilizer on seedbed was more economical than on fields.

Not all farmers can afford to procure substantial amounts of fertilizer in order to follow recommended rates in the fields. Since very few studies have been conducted regarding fertilization of rice in the seedbeds, this study was designed to evaluate the effects of nursery fertilization on the growth and yield of lowland rice.

MATERIALS AND METHODS

Description of soil used. The experiment was conducted at the Central Experiment Station, U. P. College of Agriculture, on an alluvial soil mapped as Calumpang clay. At the start of the study, the analysis of the soil showed a pH of 6.2, an organic matter content of 2.81 per cent, available phosphorous (Truog) of 4.22 ppm. and a potassium content of 0.63 m.e./100 g. of soil.

Rice variety. Milfor 6 (2), which is early-maturing, resistant to lodging, of low tillering capacity, and a fair yielder, was used as the test crop.

Seedbed preparation. Two methods of seedbed preparation were employed. These are the "dapog" and the "punla" methods. The former is usually prepared by laying banana leaves over a small area in the field. A thin layer of mud is then spread over the leaves on which previously soaked rice seeds are sown. In the "punla" method, the seeds are sown directly into the thoroughly puddled seedbed. In both methods, seeds are soaked in ordinary tap water for about 24 hours and allowed to germinate before sowing.

Forty-eight seedbed plots were laid to accommodate six treatments of four replications for each seedbed preparation. Each plot had an area of 0.84 sq. m.

The amount of rice seeds of Milfor 6 (2) used for each plot was approximately 200 g. (4000 seeds). This was found to be sufficient to transplant an area of 30 sq. m. at a distance of 20 x 40 cm. with four to five seedlings per hill.

Fertilization of the seedbed. The rates of fertilizer treatment of the seedbeds are shown in table 1. The nitrogen was supplied as ammonium sulfate (20 per cent N), phosphorus as superphosphate (20 per cent P_2O_5, and potassium as muriate of potash (60 per cent K_2O).

Drying of the seeds and/or poor growth of the seedlings in cases of good germination were observed when all the required fertilizer was applied at one time. Thus, applications were made in three installments prior to transplanting. With the "dapog" grown seedlings, the first application was done soon after sowing, the second, five days after, and the third, 10 days after sowing.

TABLE 1.—*The effect of seedbed fertilization of "dapog" seedlings of Milfor* 6 (2)

N-P_2O_5-K_2O TREATMENT	AVE. HEIGHT AT MATURITY				AVE. NUMBER OF TILLERS				AVE. NUMBER OF PANICLES			
	59 D	59 W	60 D	60 W	59 D	59 W	60 D	60 W	59 D	59 W	60 D	60 W
kg./ha.			*cm.*									
0- 0- 0	110.6	136.9	119.2	124.4	11.1	11.6	12.5	14.1	11.1	10.9	11.2	14.1
60- 0- 0	111.9	139.1	118.3	126.6	10.1	12.9	12.2	15.7	10.1	12.7	10.5	15.7
0-60- 0	112.1	142.4	114.3	119.4	10.7	14.0	12.1	15.2	10.7	13.9	10.3	15.1
0- 0-60	113.5	141.4	119.7	121.4	11.9	14.5	12.0	13.3	11.9	14.2	10.7	13.3
60-60- 0	110.2	143.0	115.2	123.8	11.8	14.0	12.9	15.8	11.8	13.6	10.4	15.8
60- 0-60	118.5	143.8	119.5	118.5	12.3	14.1	12.0	16.0	12.3	13.7	11.6	15.6
M E A N	112.5	141.1	117.7	122.3	11.3	13.5	12.3	15.0	11.3	13.4	10.9	14,9
HSD (.05)	ns[a/]	ns	ns	ns	1.81	ns	ns	ns	ns	ns	ns	ns
C.V. (%)	4.5	3.6	4.2	4.7	30.1	11.2	17.1	12.0	10.6	11.7	22.7	11.4

a/ ns — not significant

"punla"-grown seedlings received fertilizer at eight-day intervals from the date of sowing.

Transplanting and cultural operations. As was observed in previous studies that uneven maturity of grains frequently resulted in great bird damage, the seedlings raised under the "dapog" and "punla" methods were transplanted at the same time. Accordingly, the sowing of seeds for the "dapog" method was staggered by a few days so that the transplanting could be made at the same time with the "punla" seedlings. The "dapog" seedlings were about 12 to 15 days old when they were transplanted to the paddies and the "punla" seedlings, 25 to 30 days old. The plots in which the seedlings were transplanted were not fertilized.

The plants were sprayed with Endrin two to three weeks after transplanting, at about mid-tillering stage, and finally before heading. Handweeding was done whenever necessary. At harvest, the grain yield per plot, straw-weight of 10 randomly selected hills, and panicle and tiller counts were collected. The experiment was repeated for two years, making a total of four croppings. The respective planting and harvesting dates for each season were:

Cropping Season	*Date of Planting*	*Date of Harvesting*
1959 Dry	December 9, 1958	March 31, 1959
1959 Wet	July 21, 1959	October 20, 1959
1960 Dry	February 6, 1960	May 23, 1960
1960 Wet	July 20, 1960	November 1, 1960

RESULTS AND DISCUSSION

Effects of fertilization on the panicle and tiller counts and on plant height at maturity. Tables 1 and 2 show the summary data of the different growth characters studied. No significant differences in the heights of the plant at maturity which can be attributed to treatments were observed. In most instances, however, fertilized plants were slightly taller than the unfertilized ones. In addition, the mean heights of the plants varied from one season to another but they were generally taller during the wet season.

The number of tillers among treatments did not vary significantly and/or consistently in both seedbed preparations. The number of panicles did not show any significant variation due to treatments, except in one instance during the 1960 wet season

TABLE 2.—*The effect of seedbed fertilization of "punla" seedlings of Milfor 6 (2)*

N-P_2O_5-K_2O TREATMENT	AVE. HEIGHT AT MATURITY				AVE. NUMBER OF TILLERS				AVE. NUMBER OF PANICLES			
	59 D	59 W	60 D	60 W	59 D	59 W	60 D	60 W	59 D	59 W	60 D	60 W
kg./ha.			*cm.*									
0- 0- 0	116.1	129.9	118.0	121.2	11.2	9.9	12.0	13.5	10.9	9.2	8.8	13.1
60- 0- 0	116.3	127.3	117.3	126.2	10.9	10.6	11.6	15.1	10.8	10.2	8.9	14.6
0-60- 0	111.9	130.7	127.3	117.7	9.4	12.7	11.9	12.0	9.3	11.6	8.9	11.9
0- 0-60	115.3	132.2	127.8	120.3	10.2	11.8	15.0	12.8	10.0	11.6	12.2	12.4
60-60- 0	114.8	130.7	120.6	127.1	10.4	10.6	10.7	16.6	9.5	10.5	8.9	16.5
60- 0-60	113.2	130.2	119.4	125.2	10.3	11.7	11.2	14.2	10.3	11.0	8.8	14.1
M E A N	114.6	130.2	121.7	122.9	10.4	11.2	12.1	14.0	10.1	10.7	9.4	13.7
HSD (.05)	ns a/	ns	ns	7.4	ns	ns	ns	3.2	ns	ns	ns	4.5
CV (%)	2.9	3.2	4.7	2.9	16.4	19.3	20.7	9.3	14.9	17.9	31.4	9.5

a/ ns — not significant.

cropping under the "punla" method. During this season, it was seen that there were more panicles when both nitrogen and phosphorus were added in the seedbed. This trend of result was also illustrated in the height of the plants under the same method and season. The evidences obtained showed that these responses were due to nitrogen.

Effects on grain yield. A summary of the average grain yields (15 per cent moisture) during the four cropping seasons is shown in table 3. The combined analyses of variance of these data are presented in table 4. As indicated in table 3, the HSD values

TABLE 3.—*The effects of fertilizer treatments in the seedbed on grain yields in different seasons*

TREAATMENTS	1959 Dry		1959 Wet		1960 Dry		1960 Wet		MEAN
	DAPOG	PUNLA	DAPOG	PUNLA	DAPOG	PUNLA	DAPOG	PUNLA	
kg./ha.				*cav./ha.*					
0- 0- 0	61.2	55.7	78.0	62.2	54.7	34.6	64.4	53.3	58.0
60- 0- 0	64.6	64.3	101.8	85.8	51.9	32.9	76.8	60.9	67.4
0-60- 0	58.6	54.6	95.9	67.7	67.1	38.7	64.1	55.7	62 8
0- 0-60	61.9	59.8	86.4	103.7	75.6	36.9	65.5	54.6	68.0
60-60- 0	60.4	53.3	95.9	67.7	57.9	33.8	78.9	62.9	63.8
60- 0-60	68.3	57.9	86.9	104.7	64.8	31.1	74.4	59.8	68.5
M E A N	62.5	57.6	90.8	81.9	61.9	34.8	70.7	57.9	64.8
HSD (.05)	5.6	ns[b/]	20.6	ns	ns	ns	10.9	ns	8.57
SEASON MEAN [a/]	60.0		86.4		48.3		64.3		

a/ HSD (.05) between season—4.34.
b/ ns — not significant.

TABLE 4.—*Combined analyses of variance*

SOURCE OF VARIATION	D.F.	M. S.
Replication (R)	3	50.58
Season (S)	3	12,175.91 a/
R x S	9	46.39
Method (M)	1	8,731.81 a/
M x R	3	70.52
M x S	3	1,149.90 b/
M x S x R	9	281.65
Treatment (T)	5	523.67 a/
T x S	15	282.39 b/
T x M	5	125.87
T x M x S	15	312.63 a/
Pooled Error	120	139.90
Total	191	——

a/ Significant at the 1 per cent level.
b/ Significant at the 5 per cent level.

for each season show a significant response by the "dapog" seedlings during the 1959 dry, 1959 wet, and 1960 wet croppings. The yield increase was mainly due to nitrogen and an increase due to potassium addition was suggestive. The "punla"-raised seedlings showed no significant increase in yield. The lack of consistency in the response of "dapog" and "punla" seedlings may be explained by the method employed in the seedling culture. The seedbed of the "dapog" method is provided with a receptable which may insure the positional availability of the added nutrients. In the "punla" method, the added nutrients may move freely and may not be positionally available. Reyes *et al.* (1961) estimated the annual soil losses through leaching in paddy soils of essential constituents in the following magnitude: N, 4 kg.; P, 0.046 kg.; and K, 22 kg. per hectare.

The combined analyses of variance showed that the effects due to fertilizer treatments over all seasons and methods of preparation were highly significant. The mean values can be seen in the last column of table 3. All fertilized plots showed higher yields as compared with the control. The responses were mainly due to the application of nitrogen and/or potassium in the seedbeds. The result on the response of crop to potassium was particularly interesting because previous studies (Galvez, *et al.* 1956) have not shown any consistent result. It appears from this result that addition of potassium in the seedbeds may encourage better growth and consequently bigger yields.

Table 3 also reveals yield level differences among seasons. Yields obtained in the wet season were higher than those in the dry season. However, yields during a particular season varied from year to year.

Greater yields were obtained from the "dapog"-raised than from the "punla" seedlings. The faster recovery of the "dapog" seedlings compared to the "punla" may partly account for this difference. In transplanting the "punla" seedlings, the roots may have been broken during pulling and the seedlings, topped, as is usually the case. This practice invariably prolonged the period of recovery of the "punla."

The method x season and season x treatment interactions were found to be significant. The method x season interaction is shown by the trend of results given in table 5. The differences between

TABLE 5.—*The effect of season on the grain yield of "punla-" and "dapog"-raised seedlings over all treatments* a/

METHOD	1959 Dry	1959 Wet	1960 Dry	1960 Wet	METHOD MEAN
			cav./ha.		
Dapog	62.5	90.8	62.0	70.7	71.5
Punla	57.6	81.9	34.6	57.9	58.0
Season Mean	60.1	86.4	48.3	64.3	64.8

a/ Average of four replications and six treatments.

the methods of preparations were not consistent in all seasons. Table 6 shows that the treatment differences between treatments markedly varied with season and method of seedbed preparation.

TABLE 6.—*Seasonal difference between the grain yields of "dapog"- and "punla"-raised seedlings* a/

TREATMENT N-P_2O_5-K_2O	1959 Dry	1959 Wet	1960 Dry	1960 Wet	TREATMENT MEAN
kg./ha.			*cav./ha.*		
0- 0- 0	5.5	15.8	20.1	11.1	13.1
60- 0- 0	0.3	16.0	19.0	15.9	12.8
0-60- 0	4.0	28.2	28.4	8.4	17.3
0- 0-60	2.1	—17.3	38.7	10.9	8.6
60-60- 0	7.1	28.2	24.1	16.0	18.9
60- 0-60	10.4	—17.8	33.7	14.6	10.2
Season Mean	4.9	8.9	27.3	12.8	13.5

a/ Value given is "dapog" minus "punla" yields.

Effects on the straw yields. The straw yields are shown in table 7. In general, there were no differences due to treatment in

TABLE 7.—*The influence of seedbed fertilization on the straw production of the "dapog" and "punla" grown seedlings in four cropping seasons* a/

TREATMENTS N-P_2O_5-K_2O	1959 Dry		1959 Wet		1960 Dry		1960 Wet	
	Dapog	Punla	Dapog	Punla	Dapog	Punla	Dapog	Punla
kg./ha.						*tons/ha.*		
0- 0- 0	3.9	4.2	4.5	3.5	7.5	10.2	4.3	3.8
60- 0- 0	3.7	4.3	6.1	4.1	6.0	9.0	5.2	4.3
0-60- 0	3.7	3.4	6.7	4.4	6.5	9.7	4.8	4.0
0- 0-60	4.1	3.7	7.4	4.6	8.8	8.7	4.2	4.0
60-60- 0	3.8	3.5	6.3	4.4	8.5	9.1	5.1	4.4
60- 0-60	4.1	3.6	6.4	4.2	7.4	8.7	4.9	4.1
M E A N	3.9	3.8	6.2	4.2	7.5	9.2	4.8	4.1
HSD (.05)	nsb/	ns	2.2	ns	ns	ns	ns	ns
C.V. (%)	11.8	14.2	15.6	21.1	27.7	16.3	8.5	15.4

a/ Average of four replications.

b/ ns — not significant.

both the "dapog" and "punla" methods of seedbed preparation. However, in the 1959 dry season, the "dapog" showed a slight increase in straw yield due to fertilizer treatments. A similar increase was not observed in the other method. It can also be noticed that during the 1960 dry season for both methods, relatively more straw yields were obtained. However, grain yields for this season were the least as compared to any season. The coefficient of variation during this season was rather high.

The wet season of 1959 under the "dapog" method showed a slight increase in straw yield from potash application. The same trend of results was also observed in the previous season.

SUMMARY

Studies on fertilization of rice seedlings, Milfor 6 (2), raised under two methods were conducted for two years (two dry and two wet seasons) in a total of four croppings in order to determine the effects of this practice on the growth and yield of the seedlings transplanted into the field. Evidence showed that there was no apparent effect of fertilizers on the different growth characters such as plant height, and panicle and tiller counts at maturity. In addition, there was no consistent response to fertilizer on straw production. Straw yields varied from season to season.

There were, however, significant increases of grain yields due to fertilizer treatments over all seasons and preparations. The effect could be attributed to nitrogen or potassium addition in the seedbeds. Phosphorus response was not consistent. From the standpoint of preparation, however, it was shown further that only in the "dapog" method were greater responses to fertilizers obtained.

Greater yields were obtained during the wet season than during the dry season.

The evidences obtained point out the advisability of fertilizing rice seedbeds especially if prepared under the "dapog" method. It is recommended that further studies be conducted to determine the magnitude of yield response to fertilization in both seedbed and field conditions. It is possible that rate of fertilizer application in the field may be reduced if fertilization is also made on the seed-

bed.

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AN EVALUATION OF SOME METHODS FOR CHARACTERIZING THE POTASSIUM STATUS OF SOME PHILIPPINE SOILS[1/]

EDILBERTO D. REYES

WITH TWO TEXT FIGURES

The availability of potassium in surface soil samples from 10 different selected locations in the Philippines was measured through the potassium uptake of ipil-ipil (Leucaena glauca, Linn.) in six croppings and through the chemical extraction of soil potassium with neutral N ammonium acetate; 0.5, 1.0, and 2.0 N boiling nitric acid solutions; and by a continuous percolation method with 0.01 N hydrochloric acid solution as extractant.

The statistical analyses of the dry matter yields of plants warranted the arbitrary classification of the soils into: a) high yielding soils (of the Isabela, Mallig, Lipa, and Tugbok series); b) medium yielding soils (of the Quingua, Guimbalaon, and Guadalupe series); and c) low yielding soils (of the Baguio, Maligaya, and Silay series). The trend in the potassium uptake of the plants did not parallel the trend of plant dry matter production on the different soils.

The correlation coefficients between the total potassium absorbed by the plants and the levels of soil potassium extracted using various chemical methods were all found to be highly significant. The best correlation was obtained with the continuous percolation method (r — 0.988).

Results of the cropping experiment also indicated that the release of potassium from non-exchangeable forms occurred only in the soils of the Mallig, Quingua, Lipa, and Maligaya series. Some of the exchangeable potassium initially present in the other soils studied were possibly reverted to non-exchangeable forms.

Soils differ quite widely not only in their total potassium content but also in their capacity to supply available potassium to plants. The capacity of soils to supply available potassium is best assessed through the potassium uptake of plants upon successive croppings. This biological method, however, usually takes time and entails much labor. Several chemical methods, therefore, have been tried as the immediate means of predicting field responses to potassium application.

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2/ The author is grateful to former Cornell Visiting Professors in Soils N. C. Brady, M. G. Cline, and L. E. Nelson for their encouragement and guidance during the progress of this study.

Exchangeable potassium has commonly been used as a measure of potassium in soils readily available to plants. It constitutes only a fraction of the total soil potassium and is generally considered to represent both the potassium held with varying degrees of electrostatic and molecular forces around the negatively-charged soil colloids and potassium in the soil solution. Attoe (1948) and Seay *et al.* (1949) have indicated significant relationship between exchangeable potassium in the soil and that absorbed by plants.

Several workers (Chandler, *et al.*, 1945; Cholston and Hoover, 1948; Ayers, 1949; and Breland, *et al.*, 1950), however, have reserved doubts on its diagnostic value since the plants took up more potassium than could be accounted for by the initial level of exchangeable potassium in the soil. Ayres (1949), Rouse and Betramson (1949), Pratt (1951), Williams and Jenny (1952), and Kolterman and Truog (1953) have tried other chemical methods for assessing the potassium supplying power of soils.

Few studies so far have ben made regarding the potassium supplying capacity of the different soils in the Philippines. This study aimed to evaluate some of the methods for characterzing the potassium status of Philippine soils.

MATERIALS AND METHODS

Soils. Samples of the surface soil were collected from 10 different selected locations in the country. Information about these soils are presented in table 1.

Greenhouse study. The bulk samples were air-dried and ground to pass through a 2-mm. sieve. Subsamples were diluted with washed Palawan sand in the proportion of one part of soil to four parts of sand. Ammonium sulfate and ordinary superphosphate were mixed at rates equivalent to 45 kg. N and 90 kg. P_2O_5 per hectare.[3/] Calcium carbonate was also added at the rate of 1 1/2 tons per hectare for every pH unit of the soil below 6.5.

Seven-kilogram portions of each amended soil-sand mixture were weighed into coal tar-painted clay pots, each pot with an inch-hole for drainage. Each pot was then fitted on a similarly painted clay pan in which the rain or distilled water for the plants were added. The treatments, consisting of the 10 soils and pure sand

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3/ Two million kilograms of air-dry soil per hectare-furrow-slice was used in the calculation.

TABLE 1.—*Description of the soils studied*

SOIL SERIES	TEXTURE a/	LOCATION	PARENT MATERIAL
Isabela	Sandy clay loam	Hacienda San Antonio San Carlos, Negros Occ.	Alluvium
Mallig	Clay	Mallig, Isabela	Alluvium
Lipa	Clay loam	U.P.C.A. Orchard College, Laguna	Shallow alluvium over volcanic tuff
Tugbok	Clay	Davao City	Igneous rock, predominantly andesitic
Quingua	Clay loam	Mabalacat, Pampanga	Alluvium
Guimbalaon	Clay loam	Cadiz, Negros. Occ.	Old alluvium
Guadalupe	Clay	Bilibid Prison Compound, Muntinlupa, Rizal	Volcanic tuff
Baguio	Clay	Baguio City	Limestone
Maligaya	Clay	Muñoz, Nueva Ecija	Alluvium
Silay	Sandy clay loam	Silay, Negros. Occ.	Alluvium

a/ By the Bouyoucos hydrometer method (Piper, 1950).

alone, were replicated five times and laid out in a randomized complete block design.

Ten seeds of ipil-ipil (*Leucaena glauca* Linn.) were planted in each pot. A month later, the seedlings were thinned to five per pot. The tops were harvested at intervals of 1 1/2 months. Six harvests were included in the study. After the sixth harvest, the soils were sampled and the plant stubs with their roots carefully washed-off of adhering soil particles.

Laboratory techniques. The harvested plant tissue samples were oven-dried at 80°C, weighed, and then ground in a Wiley mill. The potassium contents of the tissues from the first three harvests[4]/ were determined according to the method of Wolf and Ichisaka (1947). Potassium in the tissues from the subsequent

4/ W. C. Mangupag, "The potassium supplying power of the different types of Philippine soils with ipil-ipil (Leuceana gluca. Linn.) as indicator. Undergraduate Thesis, University of the Philippines, 1956. Unpublished.

harvests were extracted using the dry ashing method employed by Hood[5/] with the extracted potassium finally taken up in 0.1 *N* nitric acid solution.

For the chemcal extraction of soil potassium, the following methods were used:

1. *Extraction with ammonium acetate*—This method involves the extraction of exchangeable soil potassium using *N* ammonium acetate, adjusted to nearly pH 7.0, at a soil-to-solution ratio of 1:10 (Peech, *et al.*, 1947).

2. *Extraction with boiling nitric acid solutions*—Ten-gram samples of the soils were boiled for 10 minutes in 100 ml. of nitric acid solutions. Three concentrations of nitric acid, *viz.*, 0.5, 1.0, and 2.0 *N* were used. After cooling and filtering the mixtures, the soils were washed with 150 ml. of warm 0.1 *N* nitric acid.

3. *Extraction through continuous percolation with dilute hydrochloric acid solution*—The method described by Garman[6/] was used. Ten liters of 0.01 *N* HCl solution were allowed to percolate through a column of 10 grams of soil intimately mixed with acid-washed quartz sand. The percolate, charged with the cations, was then led through a column of cation exchanger (Amberlite IR-120) at a rate of about one liter every six hours. Potassium was recovered from the resin after the passage of the second, fifth, and tenth liter of the percolating solution by leaching 100 ml. of *N* HCl through the spent resin column followed by a final washing with 150 ml. of distilled water.

After due destruction of organic matter and ammonium ions or after evaporating off the excess acids in the resulting extracts, the extracted potassium was finally taken up in adequate amounts of 0.1 *N* nitric acid solution. Potassium was subsequently determined using the turbidimetric method of Wolf and Ichasaka (1947).

RESULTS AND DISCUSSION

Greenhouse study. Abnormal growth of the plants was observ-

5/ J. T. Hood, "The effect of different rates, sources, and methods of application of potassium upon the potassium status of some New York soils". Ph. D. Thesis, Cornell University, 1954. Unpublished.

6/ W. L. Garman, "The characterization of the release of potassium from several Ohio and New York soils." Ph.D. Thesis, Cornell University, 1954. Unpublished.

ed during the third cropping period. It was at first believed that this was due to potassium deficiency as a result of the dilution of the soils with sand. The abnormal manifestations of the plants were later ascertained to be due to a deficiency of micronutrients. Solutions containing boron, copper, iron, manganese, and zinc were therefore added every time the plants were watered, resulting in their improved growth. It was calculated that at the termination of the experiment, each micronutrient had been applied at a rate equivalent to 12 kg. per hectare.

The cumulative dry water production and potassium uptake of the plant indicator attributed solely to each soil are presented in figures 1 and 2, respectively. The correction to soil alone was predicated on the assumption that each kilogram of the sand had proportionately contributed to the growth of the plants in each soil-sand mixture and that its contribution to plant growth was not affected by its mixing wih the soil.

Statistical analyses of the dry matter yields indicated that the 10 soils highly differed in their yielding capabilities. The results warrant the arbitrary classification of the soils into: a) high yielding soils (of the Isabela, Mallig, Lipa, and Tugbok series) : b) medium yielding soils (of the Quingua, Guimbalaon, and Guadalupe series) ; and c) low yielding soils (of the Baguio, Maligaya, and Silay series)

The trend in the potassium uptake of plants on the different soils did not parallel the trend of plant dry matter production. Plants on the high yielding soils absorbed the highest amounts of potassium. On the other hand, plants on the medium yielding soils had not necessarily extracted higher amounts of soil potassium than those on the low yielding soils. Obviously, the potassium composition of the plants varied widely, most likely very much depending on the availability of potassium in the soil.

Chemical extraction of soil potassium. The levels of potassium extracted from the soils through the different methods are presented in table 2. In no case did the total potassium absorbed by the plants exceed the chemically-extractable potassium present in the original soil samples. These chemically- extracted soil potassium were found highly correlated with the total potassium in the plant tissues.

Extraction with ammonium acetate. The total dry weights of the plant tops in six croppings were found significantly correlated

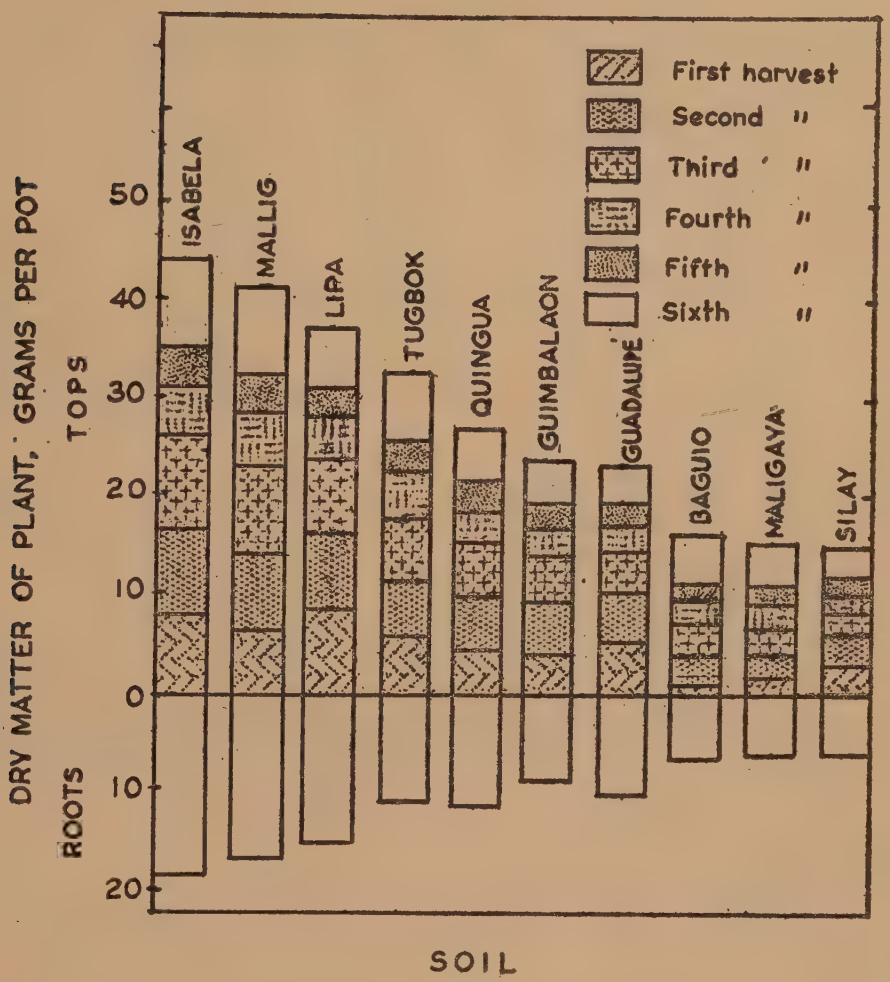

Fig. 1.—*The cumulative dry matter production if ipil-ipil* (Leucaena glauca *Linn.) in six croppings.*

with the levels of exchangeable potassium of the soils before (r—0.736) and after (r—0.882) the croppings. Likewise, the correlation coefficients betwen the total potassium in the plant tissues and the levels of exchangeable soil potassium before and after the croppings, which were correspondingly 0.928 and 0.985, were found highly significant. These would emphasize that the the relatively facile determination of exchangeable potassium is desirably adoptable in routine analysis of these soils for their fertility status.

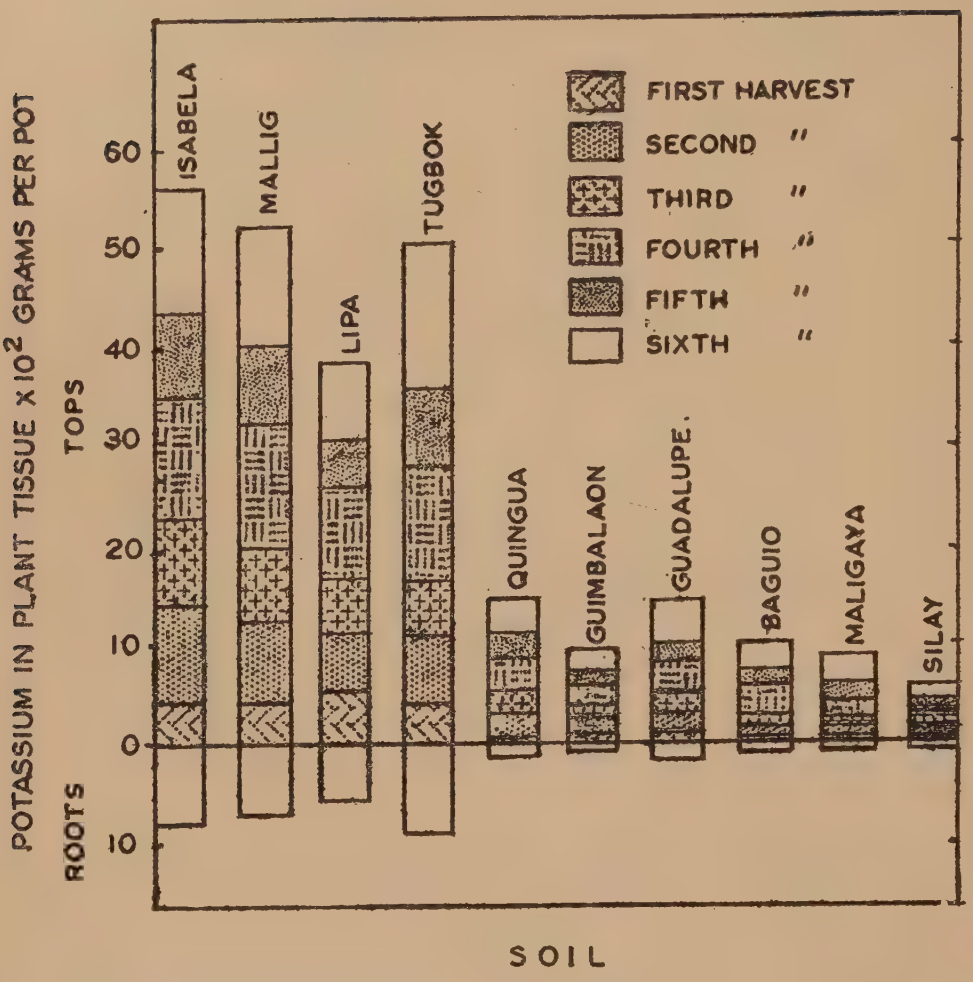

Fig. 2.—*The cumulative potassium uptake of ipil-ipil* (Leucaena glauca *Linn*). *in six croppings.*

Extraction with boiling nitric acid solutions. The amounts of potassium extracted from the soils using the three nitric acid solutions greatly exceed the levels of the readily available potassium extractable with neutral ammonium acetate. Evidently, digestion of the soils with the nitric acid solutions resulted in the extraction not only of the readily available but of the moderately available forms of soil potassium as well.

Increasing amounts of soil potassium were extracted with in-

TABLE 2.—*Total potassium extracted from the soil through cropping and different chemical methods*

SOIL	PLANT UPTAKE	CHEMICAL EXTRACTION					
		N Ammonium acetate		Boiling nitric acid			Continuous Percolation 0.01 *N* HCl
		Before Cropping	After Cropping	0.5 *N*	1.0 *N*	2.0 *N*	
		kilograms per hectare					
Isabela	914	1541	600	2180	2940	3200	2348
Mallig	814	1080	446	1280	1920	2560	1860
Lipa	622	890	323	1750	2234	2500	1723
Tugbok	838	2060	523	2440	2550	2680	2250
Quingua	220	230	80	240	600	810	640
Guimbalaon	127	160	19	275	335	340	193
Guadalupe	218	470	147	750	760	1320	732
Baguio	150	225	47	240	340	640	337
Maligaya	126	180	85	370	480	600	263
Silay	78	115	26	330	340	380	257
Correlation coefficient, r		0.928[a/]	0.985[a/]	0.925[a/]	0.970[a/]	0.977[a/]	0.988[a/]

a/ Significant at the 1 per cent level.

creasing concentrations of the acids. During the analysis, higher amounts of sesquioxides were likewise being extracted from the acid-boiled soils as evidenced by the deepening color intensity of the filtrates with higher concentrations of the acid. This would provide indirect proof of the drastic effect of the acid on the relatively weatherable potassium-bearing silicate minerals in the soils.

The amounts of potassium extracted from the soils with each nitric acid solution were all found highly correlated with the total potassium in the plant tissues. Ayres (1949), using 1.0 *N* hydrochloric acid solution on Hawaian sugar cane soils, and Rouse and Betramson (1949), using 1.0 *N* nitric acid solution on several Indiana soils, have indicated the significant relationship between the acid-extracted and plant-absorbed potassium.

Extraction through continuous percolation with HCl solution. The total amounts of the soil potassium extracted through continuous percolation of dilute hydrochloric acid solution gave the best correlation (r—0.988) with the total potassium taken up by plants.

The results of the serial analyses for soil potassium extracted through the percolation of the acid solution are presented in table 3. Except from the soil of the Guimbalaon series, the first two liters of the percolates extracted from the soils more potassium than did the ammonium acetate solution. The subsequent passage of more

TABLE 3.—*The potassium extracted from a 10-gram sample of soil through continuous percolation with 0.01 N HCl solution*

SOIL	AVERAGE POTASSIUM RELEASE		
	First 2 liters	Next 3 liters	Last 5 liters
	x 102 milligrams K per liter solution		
Isabela	447	50	26
Mallig	378	30	17
Lipa	352	25	17
Tugbok	534	9	6
Quingua	118	12	10
Guimbalaon	31	4	5
Guadalupe	137	11	11
Baguio	69	6	3
Maligaya	48	6	4
Silay	36	9	6
Correlation coefficient, r, with K uptake of plants	0.974 a/	0.764 b/	0.735 b/
Correlation coefficient, r, with exchangeable K after cropping	0.972 a/	0.773 a/	0.730 b/

a/ Significant at the 1 per cent level.
b/ Significant at the 5 per cent level.

0.01 *N* HCl solution elicited the release of lesser amounts of potassium. The results obtained by Ayres (1949) on electrodialyzed Hawaiian soils similarly suggested that exchangeable potassium is easily replaceable, requiring only small amounts of energy. Increasingly more and more energy is required for the subsequent release of potassium undoubtedly from the non-exchangeable forms.

The high yielding soils—Isabela, Mallig, and Lipa—were maintaining higher levels of replaceable potassium than the other soils after the passage of two liters of solution. The subsequent amounts of potassium released from these three soils tended to decrease while those of the other soils were lower but more constant. The high yielding soils were therefore releasing potassium from non-exchangeable forms at a faster or higher rate than the medium and low yielding soils, following the postulated equilibrium between the non-exchangeable, exchangeable, and soil-solution potassium. Apparently, the passage of five liters of solution through the three high yielding soils had not reduced the replaceable potassium level

to a commensurate measure of the characteristic level of potassium of these soils. This level, which is unique of a soil, appears to be low in the medium and low yielding soils.

The results suggest a high reliability in the method when predicting variations in the capacity of the soils to supply available potassium to plants.

Release of potassium from the non-exchangeable forms. The amount of soil potassium derived by the plants from the non-exchangeable forms was determined using the equation:

> Amount of K from non-exchangeable form = (Total K in plants plus exchangeable K in soil after cropping) minus (Exchangeable K in soil before cropping).

It was noted that the release of potassium from the non-exchangeable forms occurred only in the soils of the Mallig, Quingua, Lipa, and Maligaya series. The appreciable amount of non-exchangeable potassium rendered available to plants in the soil of the Mallig series was equivalent to 205 kilograms per hectare. This points to a fairly rapid replenishment of the exchangeable potassium from the non-exchangeable forms in this soil. There was only little conversion of non-exchangeable potassium to exchangeable forms in the soils of the Quingua, Lipa, and Maligaya series with the amounts of potassium released equivalent to 70, 55, and 31 kilograms per hectare, respectively.

Similar calculations for the remaining six soils indicated that part of their exchangeable potassium initially present must have been reverted to non-exchangeable forms. It should be mentioned that a general increase in the pH's of the soil-sand mixtures was noted at the termination of the cropping experiment. The Palawan sand used in the experiment must be of a calcareous nature and thus may have affected to some extent the uptake of potassium by plants as well as have caused the conversion of exchangeable potassium to the non-exchangeable forms. Bower and Pierre (1944) noted that the high concentrations of calcium and magnesium in a high lime soil repressed potassium absorption by different crops. Attoe (1946) concluded that one of the two types of potassium fixation in moist soil was enhanced by liming. Standford (1947) concluded that strongly absorbed ions, such as hydrogen, iron, and/or aluminum, must be removed possibly by the addition of sodium hydroxide, calcium hydroxide, and phosphate salts before potassium can be fixed by micaceous minerals. The levels of exchangeable

calcium in the soil-sand mixtures either before or after the cropping experiment, however, were not determined.

Appreciable potassium fixation occurred in the soils of the Tugbok and Guadalupe series with 34 and 23 per cent of the exchangeable potassium initially present unaccounted for by the chemical analyses of the soils and the plant tissues. Relatively small amounts of potassium were rendered non-exchangeable in the soils of the Isabela, Guimbalaon, Baguio, and Silay series. The plants on these soils were practically utilizing only the potassium in the exchangeable forms.

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THE EFFECT OF FERTILIZERS ON THE YIELD OF COFFEE IN MATUTUM, COTABATO 1/

DIOSDADO A. CARANDANG

WITH ONE TEXT FIGURE

A fertilizer experiment on coffee was conducted in Matutum, Cotabato from 1956 to 1960. The addition of potassium decreased the yield during the first two harvests. A significant increase in yield due to nitrogen was obtained during the first harvest. The three harvests markedly showed seasonal variation in yield.

The growth of the coffee industry in the Philippines depends on the improvement of such management practices as pest and disease control, prunning, and fertilization. Unfortunately, studies on these practices, as well as on the other phases of coffee culture in the Philippines are limited. Fertilization, particularly, has received little attention from agriculturists.

Findings in foreign countries show differences in fertilizer requirements of coffee. These are usually attributed to varying soil and climatic factors. In Hawaii, for instance, Goto and Fukunaga (1956) stated that a fertilizer formula of 10-5-20 applied at the rate of 1500 to 2000 pounds per acre was favorable for coffee. They also found out that a supplementary application of 500 to 800 pounds of ammonium sulfate per acre for high-yielding coffee was necessary. In the same country, Dean and Beaumonth (1938) obtained the lowest yield of coffee when no potash was applied. In Puerto Rico, where coffee is grown at higher elevations and where the climate is similar to that of the Philippines, Vicente-Chandler *et al.* (1959) recommended the application of 10-5-15 fertilizer at the rate of one ton per acre to be applied three or more times a year. If chemical analysis of coffee pulp as reported by Castro (1960) is used as the basis for determining the fertilizer need of coffee, potash would rank first, followed by nitrogen, and then by phosphorus.

In the Philippines, Tuazon and Mendoza (1960), in their experiment in Candelaria, Quezon, found appreciable increases in yield

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2/ The author is grateful to Messrs. Pedro Changco, Jr. and Valentin de la Cruz of the Matutum Development Corporation for their assistance and cooperation in the conduct of this experiment.

when 600 grams of 12-12-12 fertilizer per tree were supplemented with 120 grams of ammonium sulfate.

Experiments have been conducted in some places in the Philippines to assess the fertilizer requirement of coffee under different soil and climatic conditions. The present report deals with the results obtained in Matutum, Cotabato, where experiments were conducted from 1956 to 1960.

MATERIALS AND METHODS

The experimental area has an elevation of about 1800 feet above sea level. The climate is classified as of the fourth type (Cruz and Mendoza, 1952) and is characterized by an almost even rainfall distribution throughout the year, with an average of 15 to 20 rainy days per month. Dense clouds usually cover the area early in the afternoon, making it possible to grow coffee in the absence of permanent shade.

The soil is a loose and friable sandy loam which contains a high amount of organic matter. The chemical properties of the soil at the start of the experiment are shown in table 1.

TABLE 1.—*Chemical analysis of the soil*

DEPTH	pH	O.M.	P (TRUOG)	CEC	EXCHANGEABLE CATIONS			
					K	Ca	Mg	Mn
inches		*per cent*	*ppm*		*m.e./100 grams soil*			
0-6	5.22	8.92	103.7	23.0	0.308	5.55	2.45	0.075
6-12	5.40	4.94	31.2	15.9	0.174	3.24	2.16	0.055

The Kona variety of arabica coffee was planted in June, 1956 and crotalaria (*Crotolaria sp.*) served as temporary shade. Before planting, the coffee seedlings were pruned at the top, leaving 3 to 4 pairs of leaves. This was done to induce the growth of more verticals. The seedlings were planted at a distance of 3 1/2 meters between rows and 2 1/2 meters between trees in the row. At the time of planting, all trees were fertilized at the rate of 20 grams of ammonium sulfate per tree. Succeeding applications were 40, 40, and 60 grams of ammonium sulfate at 2, 4, and 6 months after planting, respectively. From the 8th to the 18th month after planting, 2/3 of the amount of fertilizer based on the different treatments (table 2) was applied every two months. After the 20th month, the whole amount specified in the treatment was applied an-

TABLE 2.—*Annual yield of coffee cherries*

TREATMENT a/	CROP YEAR		
	1958	1959	1960
		kg./tree	
1. Control b/	1.417	5.758	2.366
2. 100- 0- 0	2.309	6.140	2.276
3. 100-50- 0	2.847	5.807	2.067
4. 100-50-200	2.069	4.853	2.098
L.S.D.	0.769 c/	0.784 d/	N.S.
C.V. (%)	19.44	6.20	20.45

a/ Kg. of N, P205 and K20 per hectare, respectively.
b/ Fertilized with 100 grams of 12-7-20, 20 months after planting.
c/ At the 10 per cent level.
d/ At the 5 per cent level.

nually. In all cases, fertilizer was applied in band around the trees approximately under the spread of the crown.

The treatments were arranged in a randomized complete block design and replicated four times. Ten trees comprised a plot.

All plants were regularly sprayed with insecticides and fungicides six times a year, from April to December.

RESULTS AND DISCUSSION

General observations. In 1957, there was a drought which severely affected the experimental plants. The control plants suffered heavily. They became yellow, stunted in growth, and heavily defoliated. It was believed that fertilizer addition could save them so 100 grams of 12-7-20 fertilizer was applied to each tree every two months in keeping with plantation practices. This application was made 20 months after planting. After six months, the plants were able to recover although they were still smaller and had poorer growth than the treated ones.

In spite of the regular sprayings with fungicides, all the experimental plants were severely infected by coffee rust (*Hemileia vastatrix* B. et Br.). However, common pests found on most coffee plantations were not rampant. In 1961, the most prevalent pest was the Pachyrrhynchid beetle.

Effect of fertilizer on yield of cherries. The first harvest (1958) showed a significant difference in yield between treatments 1 and 2 (table 2). This could be attributed to the effect of

nitrogen, since no fertilizer was added to the control or treatment 1 from the 8th to the 18th month after planting. The fertilizer added at the 20th month (December, 1957) could not have affected the 1958 yield because the volume of crop is largely determined by the growth made in the preceding growing and crop season (Beaumont, 1939). Since the fertilizer was applied at almost flowering time, it could not have affected the growth of the bearing laterals for that year. There was no significant response to phosphorus in addition to nitrogen (treatments 2 and 3), although a higher yield was obtained when it (phosphorus) was added. The addition of potassium with nitrogen and phosphorus decreased the yield significantly at the 10 per cent level. It seemed that there was enough potassium in the soil available for plant use so that further addition of it disturbed the balance between nitrogen and potassium. In culture solution, de Camargo (1932) showed that up to 18 months, the optimum potassium-to-nitrogen ratio for arabica is between 0.5 and unity. Perhaps the absorption of potassium, calcium, and magnesium is worth investigating. Venema (1960) concluded that yield depression caused by a potassium dressing on soils poor in potassium and magnesium was mainly due to a pronounced calcium deficiency relative to potassium and magnesium in the plant, thus disturbing the normal metabolic processes. He recommends the use of magnesium in a fertilizer trial where potassium is added, since soil analysis will not determine the sufficiency or deficiency of magnesium.

In the 1959 harvest (table 2), treatment 1 could not be considered as a control plot. Although the absence of fertilizer in treatment 1 during the previous months may have had some effect on the second harvest, response to fertilizer addition could be expected. Treatment 2 with nitrogen alone gave the highest yield. Potassium, in addition to nitrogen and phosphorus, decreased yield significantly.

The yield for 1960 (table 2) showed no significant differences among treatments. The highest yield was obtained by plants in treatment 1 followed by plants in treatment 2. Perhaps, nitrogen alone should be added at this age of the plant.

Effect of different rates of 2-1-4 *ratio.* Since extensive studies in other countries seemed to prove that complete fertilizer is needed by coffee, it was decided to include treatments of varying rates of 2-1-4 ratio as used in Hawaii. The different rates used in the experiment were 50-25-100, 100-50-200 and 200-100-044 kilograms per hectare. There were no significant differences in yield among the different rates (fig. 1). The overall yield for the three har-

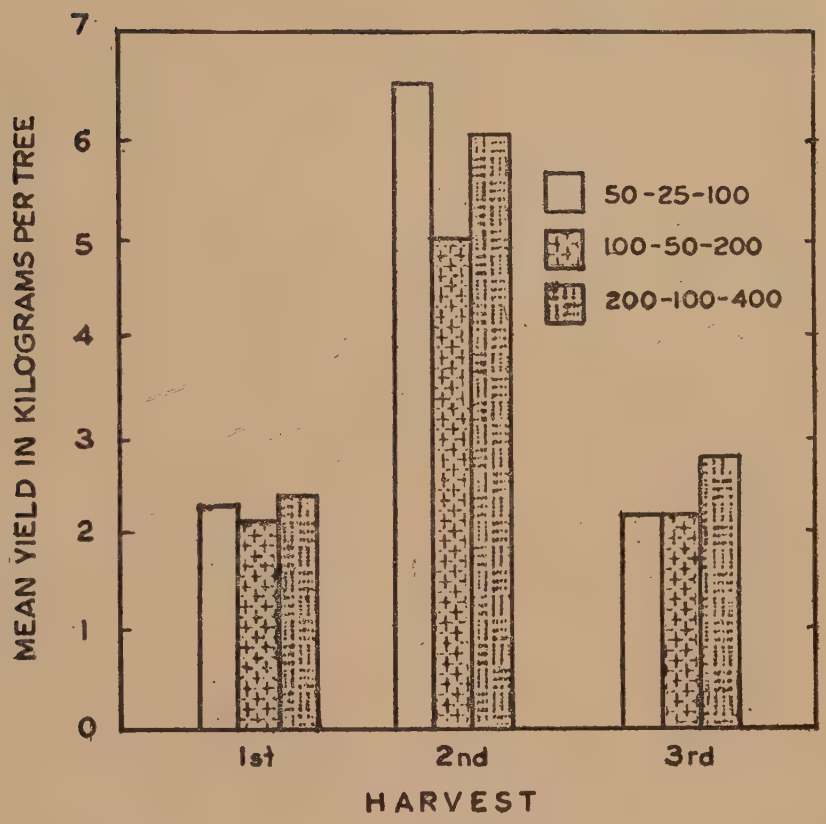

Fig. 1.—*Effect of different rates of* 2-1-4 *fertilizer ratio on the yield of coffee.*

vests showed that the lowest and the highest rates were better. It should be noted that the highest yields in all treatments were obtained during the second harvest; there was a low yield during the next harvest. This is due to the biennial bearing of coffee (Beaumont and Fukunaga, 1958).

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BIOSYNTHESIS OF C^{14}-LABELED COCONUT MEAT: II. ASSIMILATION OF C^{14}-LABELED SUGARS AND PHOTOSYNTHESIS OF GREEN NUTS [1/]

A. R. CARAÑGAL, JR. and C. R. BARIL

Excised tissue culture in d-glucose-C^{14}, injection of whole nut with C^{14}-sugar solution, and photosynthesis of detached green nut in $C^{14}O_2$ were tested for the biosynthesis of C^{14}-labeled coconut meat. D-glucose-C^{14} was assimilated and transformed into other labeled products (mostly amino acids) by excised endosperm after three hours, while C^{14}-sugars, gars, injected into detached green nut, were incorporated into various substances but to a negligible extent in the oil and starch fractions.

Photosynthesis of detached green nut in the presence of $C^{14}O_2$ appeared to be the most promising method for the biosynthesis of C^{14}-labeled meat. After the photosynthetic period, all parts of the nut were labeled.

In a recent experiment, young coconut endosperm failed to incorporate radioactive sodium bicarbonate-C^{14} (Carañgal and Baril, 1961). This is probably due to the fact that conditions inside the nut are not favorable for CO_2 fixation. The CO_2 produced during respiration is not re-used but escapes through the germinal pore. This present report deals with our attempts to biosynthesize C^{14}-labeled coconut meat through culture in C^{14}-labeled sugars and photosynthesis in $C^{14}O_2$.

MATERIALS AND METHODS

Excised tissue culture. Essentially, the procedure was similar to that previously described (Carañgal and Baril, 1961) except for some minor modifications. About 18 grams of tissue slices from the endosperm of green nut and 50 ml. of fresh coconut water containing 1 uc of uniformly labeled d-glucose-C^{14} were cultured for three hours. The alcoholic extract of the washed tissue was passed through a Dowex-50 resin (H^+ form; 5 ml. bed volume) according to the method of Plaisted (1958). This column retains the amino acids but allows the passage of the sugars (including the unmetabolized glucose-C^{14}). The retained amino acids were then eluted out

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with 4 N NH_4OH. The culture medium, alcoholic extract, and eluates from the Dowex-50 column were assayed for activity.

Injection of green nuts. Two green "coco niño" nuts—one weighing 582 grams and the other, 603 grams—were used. A hole 1/2 cm. in diameter was made on the sides of each nut and 25 ml. of the coconut water was pipeted off. This was replaced by an equal volume of C^{14}-labeled sugar solution prepared by photosynthesis of abaca and ipil-ipil. The hole was covered and the cut portion sealed with paraffin. The nuts were then placed in a desiccator.

After 86 hours, one of the nuts was analyzed. The coconut water was removed with a pipet. After repeated washings, the nut was halved. The coconut meat of one portion was extracted with alcohol and the other half was dried in the oven. The coconut water, alcoholic extract, alcohol-insoluble residue, and the dried meat were assayed for C^{14} activity. The coconut water and alcoholic extract of the endosperm were fractionated on the Dowex-50 column. After 330 hours, the other nut was analyzed following similar procedures.

Photosynthesis of detached green nuts. Two green nuts weighing 573 and 550 grams were allowed to photosynthesize in the presence of $C^{14}O_2$. After 80 hours, one of the nuts was removed from the photosynthetic chamber. The coconut water was collected and the nut halved. One portion was dried in the oven. About 10 grams of the other half was extracted with 80 per cent ethyl alcohol. The various parts of the nut (epicarp, mesocarp, and endosperm), alcoholic extract, coconut water, alcohol-insoluble endosperm were assayed for C^{14} activity. The coconut water and alcoholic extract of the endosperm were fractionated on Dowex-50 (H^+ form). After 264 hours, the other nut was analyzed similarly.

Assay of C^{14} activity. Aliquots of solutions of known volume were plated on glass planchets. The activity was then determined with a Geiger-Muller end-window counter (1.4 mg./cm^2) attached to a standard scaling circuit (the Abacus GM Scaler, Model 123, Baird Atomic). The activity of solid sample was determined by placing the counter as close as possible to the sample. The activities reported are all apparent activities.

RESULTS AND DISCUSSION

Excised tissues were able to assimilate C^{14} glucose as shown by the decrease in activity of the culture medium and the activity of the alcoholic extract of the tissues (table 1). Some of these

TABLE 1.—*Culture of excised endosperm in d-glucose-C14a/*

	ACTIVITY
	cpm.
Culture solution at start	50,250
Culture solution at end	33,495
Alcoholic extract of tissue	18,000
Alcoholic eluate from Dowex-50	13,300
NH_4OH eluate from Dowex-50	4,480
Alcohol insoluble residue of tissue	trace

a/ 18 grams of tissue was cultured in 50 ml. coconut water and 1 uc of d-glucose-C^{14} for 3 hours.

assimilated C^{14} glucose had been transformed into free amino acids (NH_4OH eluate) through normal metabolic pathways. Negligible transformation into starch and oil occurred. The alcohol-insoluble residue showed traces of C^{14} activity. The oil separated by freezing (Carañgal and Banzon, 1949) and centrifugation from the alcoholic extract was not radioactive. Higher incorporation could possibly have been obtained if a higher specific activity of glucose C^{14} were used. Glucose originally present in the coconut water (Winton and Winton, 1950) decreased the specific activity of the added radioactive glucose.

With experimental conditions similar to the natural conditions during nut development, incorporation of C^{14}-labeled sugar into the coconut meat was observed in the injection experiments of the whole nut (table 2). The incorporation, however, of the C^{14} sugar was low. The residue after alcoholic extraction of the endosperm showed C^{14} activity. Fractionation of the alcoholic extract and coconut water on Dowex-50 (H^+ form) showed transformations of the sugars to other compounds such as amino acids. A higher proportion of amino acids was present in the coconut water and alcoholic extract after a longer experimental period. Aside from the lowering of the specific activity of the injected C^{14} sugars, this method required careful disinfection to prevent spoilage of the nut.

The results presented in table 3 show that not only can C^{14}-labeled coconut meat be prepared, but the entire nut can be labeled with C^{14} by photosynthesis of detached nut in $C^{14}O_2$. C^{14} activity was detected in all parts of the nut. Unlike in the experiments above, most of the activity in the coconut water was in the sugar and organic acid fractions. C^{14} activity was also detected in the

TABLE 2.—*Injection of whole nuts with C^{14}-labeled sugar solution.*

	EXPERIMENT	
	I	II
Weight of nuts, grams	582	603
Culture period, hours	86	330
C^{14} sugar solution, volume, ml	25	25
Activity at the start, cpm	246,000	286,000
Activity at the end, cpm a/	175,000	50,000
Fractionation on Dowex-50 column		
Sugars, etc., cpm	172,800	36,500
Amino acids, etc	3,300	13,750
Alcoholic extract of 10 grams of endosperm, cpm	3,000	7,500
Sugars etc., cpm	2,542	2,300
Amino acids, cpm	340	5,000
Alcohol-insoluble endosperm b/	21	54

a/ Coconut water was assayed.
b/ Direct counting of the sample.

TABLE 3.—*Photosynthesis of detached nut in carbon dioxide—C^{14}.*

	EXPERIMENT	
	I	II
Weight of nut, grams	573	550
Photosynthetic period, hours	80	264
Activity of epicarp, cpm a/	212	990
Activity of mesocarp, cpm a/	21	54
Coconut water:		
Volume, ml.	84.8	67.5
Activity, cpm	1,900	8,437
Sugars and organic acids	1,700	8,340
Amino acids, etc.	trace	trace
Endosperm, activity		
Fresh, cpm a/	28	35
Dried, cpm a/	84	167
Alcoholic extract of 10 g. sample, cpm	5,580	21,500
Sugars and organic acids, cpm	trace	10,625
Amino acids, etc., cpm	5,000	9,730
Alcoholic-insoluble residue, cpm a/	17	81
Coconut oil, cpm b/	——	31

a/ Direct assay of sample.
b/ Dried endosperm directly crushed on planchet.

alcoholic extract of the endosperm, oil, and alcohol-insoluble residue. These results suggest that photosynthesis of the nut, inasmuch as chlorophyll is present in the epicarp, plays an important role in the development of the nut.

The photosynthesis of detached nuts appears to be most promising in the biosynthesis of labeled coconut meat. Higher C^{14} activity in the nut can possibly be obtained with higher specific activity of $C^{14}O_2$, longer experimental periods, and proper selection of the nut. The distribution of C^{14} activity in the different parts of the nut should be determined on a quantitative basis. The activity can be determined as $BaC^{14}O_3$ after the digestion of the sample with Van Slyke reagent (Aronoff, 1958). With careful separation of the various parts of the nuts (brush, shell, coconut water, and endosperm) C^{14}-labeled materials would be available for studying various problems in the utilization of the nut.

The distribution of C^{14} activity in the endosperm can also be determined. The sugars, oils, amino acids, and protein fractions can be readily separated with appropriate extraction procedures.

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THE EFFECT OF MOSAIC INFECTION ON THE CARBOHYDRATE AND NITROGENOUS CONSTITUENTS OF MUSA TEXTILIS NEE [1/]

CONCHITA ALONTE-ORILLO

Mosaic virus infection affected the pattern of distribution of some carbohydrate and nitrogenous substances in the different parts of the abaca plants. Stems of diseased and healthy plants contained the highest amounts of reducing sugars, the leaves of sucrose and of total sugars, and the roots of starch. Nitrate plus nitrite nitrogen was present in the highest amount in the false stem of both diseased and healthy plants and amide nitrogen in the roots of healthy plants and in the false stems of mosaicked plants. The different parts had a comparable level of ammonium nitrogen.

Reduction in the nitrate plus nitrite nitrogen of the roots seemed to be associated with mosaic virus infection. Accumulation of starch in all parts of the plants and of reducing sugars in the leaves accompanied the virus infection. Insignificant differences were obtained in the amounts of other carbohydrate and nitrogenous substances between the various parts of healthy and mosaicked plants.

The same free amino acids were present in the leaves of healthy and mosaic-inferted plants, but the latter seemed to have higher levels of isoleucine-leucine, phenylalanine, glutamine, and lysine. Similar chromatographic patterns of free amino acids present in the leaves of abaca, canna and arrow root were obtained. Buds of Hibiscus and cotton on which Aphis gossypii are usually found have the same free amino acids as the aphids.

Most of the past studies on the mosaic disease of abaca (*Musa-textilis* Nee) deal with is symptomatology, host range, and methods of transmission. Carangal *et al.* (1955) have initiated studies on the physiological aspects of the disease by considering the effect of virus infection on the metabolism of the abaca plant. Catalase activity and organic acid metabolism have been reported. The present investigation extends such studies to include the nitrogen and carbohydrate metabolism by determining (1) the amount of these substances, (2) the effect of the infection on the level of these substances and (3) the free amino acids of one of the insect vectors and its various host plants.

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MATERIALS AND METHODS

Six-month old abaca seedlings grown under identical environmental conditions were obtained from the Economic Garden, Bureau of Plant Industry. *Aphis gossypii* were collected from buds of *Hibiscus rosa-sinensis*. Canna and arrowroot were obtained from the Department of Plant Pathology and cotton from the Department of Agronomy, U.P. College of Agriculture.

The roots, leaves, and false stems of six healthy and six mosaicked abaca seedlings were separated, weighed and refluxed with 95 per cent ethyl alcohol for 24 hours. The alcoholic extracts were analyzed for various carbohydrate and nitrogenous constituents. The residue after alcoholic extraction was dried and ground in a Wiley mill before starch determination.

Total reducing and non-reducing sugars of the alcoholic extracts were determined according to the Munson-Walker general method (A.O.A.C., 1955). Starch was analyzed in the ground residue following the methods of the A.O.A.C. (1955). Ammonium, amide, nitrate and nitrite nitrogen were determined from the extracts according to the method of Varner *et al.* (1952). Relative amounts of asparagine and glutamine in the different parts of the abaca were analyzed from the alcoholic extract (Carangal and Varner, 1959) 2/.

Paper chromatographic analysis (Block, *et al.*, 1958) were done to detect qualitatively the free amino acids (two-dimensional) of the leaves of mosaic infected and healthy abaca, canna, *Hibiscus*. arrowroot, and *Aphis gossypii*.

RESULTS AND DISCUSSION

Effect on soluble carbohydrates and starch. The distribution pattern of the total sugars in the healthy plants was the same as in the diseased plants (table 1a). The magnitude of the values follow in the order: leaves = false stem > root. There was no significant difference in the total sugar contents of the mosaicked and healthy plants.

Mosaic infection caused differences, however, in the distribution of reducing sugars and sucrose in various parts of the plants (table 1a and 1b). In healthy plants the false stem had higher reducing sugars than roots which had comparable level with the leaves. In the diseased plants, the order was stem > leaves > roots.

2/ A. R. Carangal and J. E. Varner, "The preparation of N^{13} labelled compounds for nitrogen metabolism studies." Paper presented at the American Society of Plant Physiologists, University Park, Pennsylvania, August 30—September 2, 1959. Unpublished.

TABLE 1a.—*The carbohydrate content of a healthy and a mosaicked abaca seedling*

CARBOHYDRATE CONSTITUENTS	PLANT PARTS	DISEASED	HEALTHY
		mg./10 g. fresh weight	
Total sugars	Root	18.40 a[a/]	20.40 a
	False stem	83.52 b	76.98 b
	Leaves	79.28 b	91.08 b
Reducing sugars	Root	13.96 a	12.58 a
	False stem	40.01 b	49.38 b
	Leaves	14.50 a	27.78 c
Sucrose	Root	4.22 a	7.43 a
	False stem	41.31 b	26.22 a
	Leaves	61.50 c	60.18 b
Starch	Root	171.43 a	542.93 a
	False stem	91.13 b	308.67 b
	Leaves	119.22 b	302.25 b

a/ Values in a particular carbohydrate constituent in a given column not followed by the same letter are significantly different.

TABLE 1b.—*Level of significance in the comparison between the carbohydrates of various parts of a healthy (H) and diseased (D) abaca seedling*

CARBOHYDRATE CONSTITUENTS	ROOT	FALSE STEM	LEAVES
Total sugars	n.s. /[a]	n.s.	n.s.
Reducing sugars	n.s.	n.s.	(H < D) [b/]
Sucrose	n.s.	(H < D) [b/]	n.s.
Starch	(H < D) [c/]	(H < D) [c/]	(H < D) [c/]

a/ n.s. — not significant.
b/ Significant at the 5 per cent level.
c/ Significant at the 1 per cent level.

In both healthy and mosaicked plants, the leaves had the highest level of sucrose followed by the false stem and then the roots. The reducing sugars in the roots and false stem did not differ significantly. Leaves of diseased plants contained more reducing sugars than the healthy plants, suggesting accumulation of these sugars in the diseased leaves. The false stem of mosaicked plants had less sucrose than that of the healthy ones. There were no significant differences in the amount of sucrose in the roots and leaves of the healthy and diseased plants.

In both diseased and healthy seedlings, the roots contained the highest amount of starch while the false stems and leaves had almost the same amount. A comparison of the starch content (tables 1a and 1b) seemed to indicate that mosaic infection caused accumulation of starch in all parts of the seedlings. It is also probable that such starch accumulation in mosaicked seedlings is the net effect of virus infection on the enzymes (amylase, phosphorylase and phosphatase) involved in starch metabolism. Differences in enzymatic activities were observed by Gerola and Vaccari (1955) in their work with virus-infected tobacco.

Starch accumulation in plants affected by virus was also reported in such diseases as potato leaf roll, "spike" of sandal, spinach blight, peach yellows and little peach (Smith, 1951). Quanjer (1913) considers that this accumulation of starch is due to phloem necrosis, but Murphy (1923) and Bawden (1932) consider such phloem necrosis as secondary to the starch accumulation. Cytological studies may show whether necrosis is related to the high level of starch in the roots of abaca.

The accumulation of reducing sugars in the leaves of mosaic-infected plants is in agreement with Schweizer's (1930) finding that leaves of leaf roll-infected potato had more copious accumulation of glucose which extended to the tubers. However, in the case of spinach blight, Hawkins, as cited by Smith (1951), reported that levels of total sugars and starch were practically alike in healthy and diseased plants. Mosiac-infected abaca plants had comparable levels of total sugars and higher starch than healthy plants. A study of several mosaic disease of plants by Dunlap (1930) showed that infection by these diseases was accompanied by a decrease in the total carbohydrate content of the foliage. In the light of these findings on various types of virus disease on the carbohydrate content of the host plants. The changes in amounts of simple sugars and starch are characteristics of some but not all of virus diseases.

Effect on nitrogenous constituents. In both healthy and diseased plants, the ammonium nitrogen content of the various parts of the plant did not differ significantly (table 2a and 2b). Comparison

TABLE 2a.—*Nitrogenous substances in a healthy and a mosaicked abaca seedling*

NITROGENOUS SUBSTANCES	PLANT PARTS	HEALTHY	DISEASED
		meq. x 10^2/100 g. fresh weight	
Ammonium nitrogen	Roots	1.48 a/	2.07 a
	False stem	2.17 a	3.02 a
	Leaves	1.77 a	2.41 a
Nitrate plus nitrite nitrogen	Roots	36.71 a	7.93 a
	False stem	46.24 b/	16.05 b
	Leaves	6.78 c	8.03 c
Amide nitrogen	Roots	13.88 a	3.57 a
	False stem	5.90 b	9.89 b
	Leaves	4.75 b	5.39 a

a/ Values in a particular constituent in a given column not followed by the same letter are significantly different.

TABLE 2b.—*Level of significance in the comparison between the nitrogenous substances of various parts of a healthy (H) and diseased (D) abaca seedling*

NITROGENOUS SUBSTANCES	ROOTS	FALSE STEM	LEAVES
Ammonium nitrogen	n.s. a/	n.s.	n.s.
Nitrate plus nitrite nitrogen	(H > D) b/	(H > D) b/	n.s.
Amide nitrogen	(H > D) b/	n.s.	n.s.

a/ n.s. — not significant.
b/ Significant at the 1 per cent level.

of the corresponding parts of the normal and mosaicked plants also showed no difference. The false stem of healthy plants had the highest level of nitrate plus nitrite nitrogen, followed by the roots, and then by the leaves which had the least. In mosaicked plants, the roots and leaves had the same amounts, both of which were lower than that of the false stem. The roots and false stems of a healthy seedling had a significantly higher level of nitrate plus nitrite nitrogen than the corresponding part of a diseased seedling. The difference in amount in the leaves was not significant.

The highest amount of amide nitrogen was present in the roots

of healthy seedlings and in the false stems of mosaicked seedlings. The levels of amide nitrogen were the same in the other parts of the plants. The amide nitrogen content of the roots of a healthy plant was significantly higher than that of a mosaicked plant while those of the other parts were not. Most of the amides present in the different parts of an abaca plant was asparagine rather than glutamine (table 3).

TABLE 3.—*Determination of the relative amounts of glutamine and asparagine amide nitrogen in healthy abaca seedlings*

PARTS OF SEEDLINGS	TRIALS	GLUTAMINE	ASPARAGINE
		milliequivalents x 10^2	
Roots	I	0.25	1.30
	II	0.30	1.52
	III	0.22	1.72
	Mean	0.26	1.51
False stem	I	0.48	1.08
	II	0.30	1.18
	III	0.24	1.10
	Mean	0.34	1.12
Leaves	I	0.41	3.56
	II	0.24	2.30
	III	0.34	3.25
	Mean	0.33	3.04

It would appear, therefore, that lower amounts of nitrate plus nitrite nitrogen in the roots and false stem and amide nitrogen in the roots (table 2a) may be associated with the mosaic virus infection. The nitrogenous constituents of the leaves, which show the characteristic mosaic symptoms, were not affected by virus infection. The lower values of nitrate plus nitrite nitrogen in the false stem of the diseased seedlings appear to indicate that virus infection affects the assimilation of nitrate by the roots. Nitrite reductase activity may also be higher in those parts of the diseased plants that supply the required amino acids for the virus synthesis. Commoner

and Nehari (1953) reported that a transient deficiency develops in amides and other amino acids during rapid TMV formation.

Some of the results mentioned above are quite similar to those obtained by Jodidi *et al.* (1920) who reported that mosaicked plants have a smaller percentage of total nitrate, acid amide and mono- and diamino-nitrogen but have a larger percentage of ammonia than normal spinach plants. Nitrous acid, however, was present in the diseased plants only.

Increased asparagine formation was observed by Laloraya *et al.* (1956) in leaves of *Carica papaya* infected with carica-curl virus. Asparagine was formed in virus-infected leaves of *Hibiscus esculentus* (Govindjie *et al.*, 1956). No consistent results were obtained in the comparison of amide contents of the different parts of normal and mosaic-infected abaca plants. Humphries and Kassamis (1955) reported that only nitrate nitrogen was correlated negatively to the susceptibility of tobacco to TMV infection; amide, soluble amino acids and ammonia were not.

Free amino acids. Almost identical chromatographic patterns were found of the free amino acids of the leaves of abaca, cotton, *Hibiscus*, arrowroot and canna. There were as many as 12 separated ninhydrin positive spots which correspond to cystine, aspartic, asparagine, glutamine, glutamic acid, arginine, leucine-isoleucine, phenylalanine, tryptophan, alanine, threonine, serine, glycine and lysine according to Aronoff's chromatomap of amino acids (Aronoff, 1958).

Leaves of healthy and mosaicked-infected abaca seedlings had a similar chromatographic pattern of free amino acids. However, on the basis of the intensity of the ninhydrin-positive spots, mosaic-infected leaves appeared to have higher amounts of leucine-isoleucine, phenylalanine, glutamine and lysine. The higher level of glutamine in the mosaic-infected leaves has to be verified further.

Differences in the free amino acid composition of the leaves of healthy and virus-infected plants have also been found by various workers. Infected leaves of *Carica papaya* and *Hibiscus esculentus* contained substantially more asparagine than normal leaves (Laloraya, *et al.*, 1956). Leaves of *Croton sparsiflorus* infected with yellow mosaic virus showed the same free amino acids in increased amount and an additional prominent band corresponding to lysine-histidine (Ranjan, *et al.*, 1955). Fife (1956) also found a higher concentration of certain amino acids (arginine) in leaves of sugar

beets affected with curly top virus. Glutamic and valine were higher in juices from virus-infected potato plants than in juices from healthy plants. In contrast, Rao *et al.* (1956) reported that valine, methionine and aspartic acid were absent in virus-infected leaves of *Trichosanthes anguina* but were present in normal leaves.

Buds of *Hibiscus* and cotton leaves where *Aphis gossypii* are usually found had the same free amino acids as the abaca. However, there were no marked differences in the intensity of the ninhydrin spots, suggesting comparable levels of the different amino acids. *Aphis gossypii* had the same chromatographic pattern of free amino acids as *Hibiscus* which was to be expected since aphids ingest free amino acids and amides found in the phloem sap (Mittler, 1953).

The similarity in the free amino acid pattern of abaca, arrowroot and canna plants should not be taken into account with regard to the susceptibility of these plants to abaca mosaic virus. The free amino acids present are of common occurrence in plants.

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EFFECTS OF HIGH LEVELS OF COPRA MEAL ON THE QUANTITY AND QUALITY OF MILK OF COWS AND CARABAOS [1/]

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Cows and carabaos were fed concentrate mixtures containing 35, 45, and 55 per cent copra meal in order to compare their effects on the quantity and quality of milk produced. There were highly significant differences in the butterfat tests as the level of copra meal was increased. The fat tests increased from 3.80 per cent for cows fed a mixture containing 35 per cent copra meal to 3.97 per cent for those fed with 45 per cent copra meal and to 4.00 per cent for those fed with 55 per cent copra meal. However, the difference in the fat tests of the milk from cows fed the second and third levels of copra meal was not significant. Carabaos' milk did not show any appreciable increase in fat tests due to the increasing levels of copra meal. The four per cent fat-corrected milk, chlorides, lactose, specific gravity, iodine number, and melting points of milk from cows or carabaos did not vary signficantly.

The cost of the mixture decreased as the amount of copra meal was increased. If abundant and cheap, the use of high levels of this feed in rations for milking cows and carabaos is practical.

Copra cake, a by-product in the extraction of coconut oil from copra, is available in the Philippines throughout the year. It is also called copra meal when ground for feeding purposes. Because of its protein content of about 20 per cent, it is classified as a good source of protein and is considered as good as, if not better, than corn gluten feed for the feeding of dairy cattle. Copra cake is cheaper than either corn or fine rice bran which are the two principal ingredients in a concentrate mixture for dairy cattle.

Of the total amount of copra cake produced in the Philippines, only a small quantity is used locally by livestock raisers and poultrymen. One coconut oil factory, for instance, produces about 1,100 tons of copra cake a month but only about 50 tons of it is sold for livestock and poultry feeds. The bulk of the copra cake is exported to foreign countries, particularly to Germany. This situation implies, among other things, that our livestockmen do not fully realize the importance of this high proteinaceous feed ingredient in feeding practices.

In his study on pigs, Gallardo (1930) observed that when the

1/ Central Experiment Station Contribution No. 2829.

amount of copra cake in a ration was increased, the unpalatability of this ration also increased. He further reported that the pigs given this ration were constipated. No explanation was advanced for the pigs being so, although one would possibly expect diarrhea due to the high fat ration. Sulit (1926) fed 75 to 85 per cent copra meal to rats with resultant deleterious effects such as stunted growth, emaciation, inactivity, and weakness.

One of the criticisms against the use of high levels of copra meal is its reported deficiency in some amino acids, notably tryptophan, lysine, methionine, and histidine. (National Research Council. 1956; Bendana-Brown, *et al.*, 1958-59). However, it is possible that the presence of amino acids from other feed ingredients in a concentrate mixture would correct this deficiency. Furthermore, ruminants do not need high quality proteins because their microbial population can synthesize proteins.

Although it is impractical to feed monogastric animals with levels as high as those used by Sulit (1926a, 1926b), the use of more copra meal for ruminants may be warranted. This could be done in situations where there might be shortages of some other feed ingredients. Furthermore, the increased utilization of copra meal would undoubtedly be advantageous to both the coconut and dairy enterprises. This study was made to determine the effects of feeding high levels of copra meal on the quantity and quality of milk of cows and carabaos.

MATERIALS AND METHODS

Milking cows were selected on the bases of similarities in blood composition, stage of lactation, and milk production. Nine Holstein X Red Sindhi cows were used in a 3 x 3 Latin square design. Each feeding period consisted of six weeks for each of the three rations. In a similar manner, six carabaos (Murrah and grade Murrah buffaloes) were selected and equally distributed among the three rations. At about the middle of the second feeding period, one carabao unexpectedly dried up. Data obtained from this animal during the first feeding period were not included in the analysis of variance.

The ingredients and cost of each concentrate mixture were:

	Ration 1	Ration 2	Ration 3
	kg.	*kg.*	*kg.*
Copra meal	35.0	45.0	55.0

Yellow corn	24.0	20.0	17.0
Fine rice bran	21.0	20.0	18.0
Soybean oil meal	7.0	5.0	3.0
Corn gluten feed	11.0	8.0	5.0
Shells	1.0	1.0	1.0
Salt	1.0	1.0	1.0
Total	100.0	100.0	100.0
Calculated D. P.	15.0	15.0	15.0
Calculated T.D.N. ..	75.0	75.1	75.1
Cost of concentrate mixture	₱ 19.89	₱ 19.36	₱ 16.93

The calculation of digestible protein and total digestible nutrients was based on Morrison (1948).

The daily milk production of each animal was used as the basis for computing the amount of concentrates given to each animal. The concentrate-to-milk ratio for cows was 1:3 and for carabaos, 1:2. Every week, a two-day composite sample of the milk was analyzed for certain chemical and physical properties. The Babcock method was used for the fat tests; Munson and Walker's, for lactose determination; and Sharp and Struble's, for chloride analysis. A pycnometer was used for determining specific gravity. Butter was made from the sample and used for the determination of melting points and iodine number by the A.O.A.C.

In the statistical analysis, only data taken in the last four weeks of the experimental period were used. The first two weeks served as the preliminary feeding period.

RESULTS AND DISCUSSION

Milk production. There were no significant differences in the 4 per cent fat-corrected milk production of both cows and carabaos fed with concentrate mixtures containing 35, 45, and 55 per cent copra meal as shown in table 1. This might be due to the fact that the three mixtures had the same calculated amounts of digestible protein and total digestible nutrients. Because of the high level of copra meal, the energy content of each mixture was high. The results for cows corroborate the report of Warner *et al.* (1957), who compared mixtures with 4 per cent each of copra meal, corn gluten fed, and corn distillers dried grains for milk production. No study has been reported for carabaos or buffaloes.

TABLE 1.—*Summary of data on the quantity and quality of milk produced by dairy cows and carabaos fed with high levels of copra meal*

	Ration No. 1		Ration No. 2		Ration No. 3		Average	
	Cow	Carabao	Cow	Carabao	Cow	Carabao	Cow	Carabao
Number of animals	9	5	9	5	9	5	9	5
Individual av. da. 4% FCM Production	5.67	4.24	5.69	4.27	5.68	4.10	5.68	4.20
Butterfat test, per cent	3.80 a/	8.55	3.97 a/	8.53	4.00 a	8.53	3.92	8.54
Chlorides, per cent	0.134	0.109	0.136	0.117	0.136	0.121	0.135	0.116
Lactose, per cent	4.82	4.73	4.64	4.54	4.64	4.30	4.70	4.52
Chloride-lactose number	2.78	2.30	2.93	2.58	2.93	2.81	2.88	2.56
Iodine number	35.2	34.0	34.6	34.9	33.9	33.3	34.6	34.1
Melting point, °C	30.11	30.82	29.67	31.26	30.68	32.21	30.153	31.43
Specific gravity, 29.5°C	1.0285	1.0297	1.0284	1.0297	1.0285	1.0307	1.0285	1.0300
Amount of concentrates fed (Concentrate to milk ratio was 1:3 for cows and 1:2 for carabaos)	1.95	1.26	1.91	1.27	1.89	1.22	1.92	1.25
Ave. gain or loss in body wt., kg.	—6.81	—5.21	—0.70	6.06	—8.29	0.53	—5.27	0.46

a/ Butterfat tests for cows have highly significant differences. Using Tukey's test for significance among means, D — 0.16%.

45% copra meal (R-2) 35% copra meal (R-1) 55% Copra meal (R-3)

The effect of stage of lactation is shown in the decrease of the fat-corrected milk from 5.98 kg. in the first feeding to 5.03 kg. in the third feeding period. The corresponding increase in the butterfat tests was from 3.84 to 3.98 per cent in cows. The fat corrected milk for carabaos decreased from 4.46 kg. in the first feeding to 3.76 kg. in the third feeding. The corresponding fat tests increased from 8.42 to 8.68 per cent. The effects of stage of lactation in both species were not significant. The decrease in milk production and the increase of butterfat tests are considered normal phenomena accompanying advance in lactation.

Butterfat tests. Fat tests increased significantly ($<.01$) from 3.08 per cent for cows fed 35 per cent copra meal to 3.97 per cent for those fed 45 per cent copra meal and 4.00 per cent for those fed 55 per cent copra meal. However, the difference in the fat tests between cows fed 45 per cent copra meal was not significant (Tukey's test, D—0.16 per cent) (Snedecor, 1951). Significant increases in fat tests were also reported by Brouwer (1930), Schmidt *et al.* (1932), and Warner *et al.* (1957). Butterfat test results of this study as well as those previously mentioned are lower than those reported by Steensberg (1937) as cited by Hammond (1957), which were 0.3 per cent in the first two weeks and 0.5 per cent in the seventh to eight week of feeding. Contrary to these studies, Perkins *et al.* (1931) did not observe any increase in fat tests among cows fed copra meal.

In the case of carabaos, no significant differences in fat tests were observed in spite of the differences in the levels of copra meal fed to the animals (table 1).

Chloride, lactose, and chloride-lactose number. Chlorides in the milk did not increase significantly as the levels of copra meal were increased. This might be due to the fact that copra meal does not contain chloride. Lactose did not vary significantly as the levels of copra meal were increased. On the other hand, when lactose is low, chlorides are high. Such relationship is evident in the milk of both species (table 1). This relationship is to be expected since the mammary gland generally maintains the same osmotic pressure with blood.

Koestler (1920) suggested the use of chloride-lactose number to indicate normal milk. This number is obtained by using the following formula: $\frac{\%\ \text{chloride}}{\%\ \text{lactose}} \times 100$. The chloride-lactose number of 1.5 to 3.0 is used to indicate normal-testing milk. A high

chloride-lactose number occurs in late lactation or in mastitis milk because low lactose is accompanied by marked increase of chloride. Results of this study show that milk from cows and carabaos was normal. The average chloride-lactose number for cows was 2.88 and for carabaos, 2.56.

Iodine number. Iodine values of milkfat obtained from animals fed different levels of copra meal did not vary significantly (table 1). The values of 33.3 — 35.2 are within the range of 26 — 30 which are considered normal values as indicated by Schmidt *et al.* (1932) and Jenness and Patton (1959). Both species have almost the same iodine values of 34.6 for cows and 34.1 for carabaos.

Hill and Palmer (1938) and Brown *et al.* (1941) agree that the addition of coconut oil to the concentrate fed depresses the iodine value by about 4 to 5 units. Both of their reports mentioned the addition of 1 pound (454 grams) of coconut oil per day per cow. The depressing effect of coconut oil might be ascribed to its normal iodine values of 8 to 11.

In the present study on the other hand, the cows and carabaos did not receive as much coconut fat to cause a depression in iodine values. The cows consumed 53 to 81 grams of coconut fat every day and the carabaos, 34 to 52 grams, based on the fat content of 7.8 per cent for copra meal (Ynalvez, *et al.*, 1954). These amounts are very much lower than those reported in other studies (Hill and Palmer, 1938; Brown, *et al.*, 1941).

During the first feeding period, the iodine values were examined to find out any effect of the different levels of copra meal. This was considered necessary because the animals were not previously subjected to any of the treatment levels. A consistent increase in the iodine values for individual cows and carabaos from the first to the second week was observed. The iodine value for the cows was 30.4 for the first week and 37.2 for the second week. The latter value was the maximum attained in the entire six-week period. Thereafter, the iodine values decreased. The corresponding values for the carabaos were 27.5 and 31.8. A decrease after the second week was also observed.

Melting point. There were no significant differences in melting points as influenced by the levels of copra meal. The average for cows was 30.15°C and for carabaos, 31.43°C. Butter from carabao milk had consistently higher melting points than cows. The normal melting point values of butter are 28°C to 36°C (Jenness and Patton, 1959; Smith, 1959).

During the first feeding period, the melting points for the first and second weeks were compared. Butter from cows' milk gave a melting point of 32.02°C in the first week and 33.11°C in the second week. The corresponding melting points for butter from carabaos' milk were 33.30°C and 35.40°C. In both species, melting points decreased after the second week.

Most literatures indicate that the melting point values of coconut oil range from 20°C to 28°C. The use of high levels of copra meal did not depress the melting points of the butter produced. The small amount of coconut fat actually consumed by the animals in both species may explain the absence of depressing effects. Hill and Palmer (1938) reported that feeding of 0.8 to 1.0 pound of coconut oil per day to each cow markedly increased the hardness of butterfat.

Results of this study do not indicate that hard butter might be produced by the feeding of high levels of copra meal to dairy animals. The practical importance of hard butter in the tropics might be worth looking into because of the absence of refrigeration facilities in the majority of Filipino homes.

Specific gravity. There were no significant differences in specific gravity taken at 29°C for milk from cows and carabaos. Cows' milk had a consistent slightly lower specific gravity than carabaos' milk. The average for cows was 1.0285 and for carabaos, 1.0300. Hassan 2/ reported that individual buffalo's milk in Egypt had an average specific gravity of 1.0325 to 60°F (15.5°C).

Cost of concentrate mixture. The rations containing 55 per cent copra meal costs less than those containing 45 and 35 per cent copra meal. The cost of the mixture increased as the amount of copra meal decreased. Costwise, this may suggest that the use of more copra meal in feeds for milking animals is a practical approach. The use could be further justified in situations where some ingredients are lacking. Furthermore, levels of 35 to 55 per cent copra meal did not diminish the quantity and quality of milk produced.

Gain or loss in body weight. There were no significant differences in the gain or loss in weights in both species of dairy animals. The cows lost an average of 5.27 kg. during the experimental periods while the carabaos gained 0.46 kg. The loss in weights of cows indicates that the energy they obtained from grazing was a trifle short of the needs for maintenance and milk pro-

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2/ A. H. Hassan "The composition of milk of cows and buffaloes. Thesis, Cairo University. Unpublished.

duction. Dairy animals of both species grazed in the same kind of pasture.

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ERRATA

In volume XLV, Nos. 1 & 2, June-July, 1961 number of *The Philippine Agriculturist*, page 19, line 4 of the 6th paragraph, the sentence "But consumer differences differ". should read "But consumer preferences differ". In the same number, page 22, footnote No. 16 is "Luciano Salonga, "Monthly Review of Philippine Business Condition," Manila, 1956. Also, in the journal mentioned above, table 2b, page 32, the amount of glutamine in trial II should be 1.02.

In the *Errata* placed on volume XLV, No. 5, October, 1961 number of *The Philippine Agriculturist*, the formula of the article, "Determination of the Digestibility of Forage with Plant Chromogens as Indicator", by C. A. Mabasa, Jr., *et al.* was incorrectly printed. It should be:

$$\text{Dig. Coef.} = 100 - 100\frac{\text{(Concentraton of chromogen in forage)}}{\text{(Concentration of chromogen in feces)}}$$

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